

ing of hexamethylene diisocyanate trimer (HDI), and a hardener comprising 50 parts 900-Da polyol, 50 parts 600-Da PEG, and other essential compounds. BMP-2 powder in the presence of glucose (wt 2% in the foam) and heparin (20 fold that of protein), or PLGA microspheres was incorporated into PUR composite scaffolds through mixing with the hardener component before the foaming reaction.

[0140] The SEM image of PUR without PLGA particles is shown in FIG. 6A, and the inclusion of PLGA particles maintained the morphology property of PUR scaffold (FIG. 17). The polyurethane scaffolds contain interconnected pores with the size in the range of several hundred microns. This indicates that PUR scaffolds containing PLGA particles can serve as the matrix for cell growth and penetrating.

[0141] Release of BSA-FITC from PUR and PUR/PLGA Scaffolds

[0142] To evaluate the effect of encapsulation protein into PLGA particles on the release kinetics from PUR scaffold, small pieces of PUR scaffold incorporated with BSA-FITC powder or PLGA particles encapsulated with BSA-FITC were incubated in α -MEM containing 1% BSA under 37° C. the medium was changed as indicated in FIG. 18. The amount of BSA-FITC was determined by emission fluorescence at 530 nm after excitation at 485 nm. The BSA-FITC release profiles from PUR scaffolds shows a lower burst release by adopting the PLGA microsphere encapsulation strategy compared with directly incorporating BSA into PUR scaffold as powder, and a more sustainable release when decreasing the PLGA microsphere size from 80 μ m to 1 μ m (FIG. 18).

[0143] Release of BMP-2 from PUR and PUR/PLGA Scaffolds

[0144] Small pieces of PUR scaffold incorporated with BMP-2 powder or PLGA particles encapsulated with BMP-2 were incubated in α -MEM containing 1% BSA under 37° C., and the medium was changed every 24 hours. Immunoassay (Human BMP-2 ELISA kit, from R&D systems) was adopted to determine the amount of BMP-2 released from the PUR scaffolds. Similarly, when BMP-2 was incorporated in PUR scaffolds as a powder, a burst release occurred on the first day and little BMP-2 was release after day 8 (FIG. 19). When BMP-2 as encapsulated into large PLGA microspheres at the average size of 80 μ m and the particles embedded in PUR scaffolds, the burst release was substantially reduced and the release sustained up to 15 days (FIG. 19). Based on BSA-FITC release data, further decreasing the size of the PLGA particles is expected to achieve a more sustained profile, and the experiment is undergoing.

[0145] In Vitro Bioactivity of Released BMP-2 from PUR Scaffolds

[0146] BMP-2 is known to stimulate alkaline phosphatase (ALP) expression and mineralization of MC3T3 cells. To evaluate the bioactivity of the released BMP-2, both ALP and mineralization assays (Von Kossa staining) were carried out. Based on the release profile, PUR/BMP-2 release samples from day 1 to 8 and PUR/PLGA-L-BMP-2 from day 1 to 10 were collected for the analysis respectively. Although weaker than the positive control, the BMP-2 released from PUR scaffold stimulated ALP expression of MC3T3 cells, which verified the in vitro bioactivity of the BMP-2 in the liquid releasates (FIG. 20).

Example 7

[0147] This example helps demonstrate that biologically active molecules, including growth factors and antibiotics,

can be released from biodegradable polyurethane scaffolds, and particularly that poly(ester urethane)urea/microparticle-tobramycin (PEUUR/MP-T) composite biomaterials promote healing of infected bone wounds in an established infection model

[0148] PUR Foams were Synthesized as Shown Herein.

[0149] Evaluate the ability of PEUUR/MP-T composite biomaterials to promote healing of infected bone wounds in an established infection model. A pilot study in an infected rat segmental defect model was initiated on Apr. 7, 2008. The time point was selected as 2 weeks, and the T6C3G1L-PEG0 and T6C3G1L-PEG30 materials were tested, along with a PMMA positive control. A photograph of the defect is shown in FIG. 1. The defect was infected by placing 30 mg of type I bovine collagen wetted with 100 μ l *Staphylococcus aureus* Xen 36 in the wound (average Inoculating dose 3.30 \times 10⁵ cfu/ml) and debriding on day 14 post-infection. The test articles were then implanted into the defects and bacterial counts measured on day 14 post-implantation. Data are shown in FIG. 22.

[0150] BMP-2 was reconstituted in PBS according to the manufacturer's instructions and mixed with heparin and glucose. The resulting solution was lyophilized to yield a dry powder, which was added to the hardener component of the PUR scaffold prior to mixing. Subsequently, Desmodur N3300A polyisocyanate (hexamethylene diisocyanate trimer) was added to the hardener component to prepare the PUR scaffold using published techniques. The PUR scaffolds each contained 2.5 μ g BMP-2, 2 wt-% glucose (excipient), and 0.05 wt % heparin to stabilize the BMP-2. In some cases (PLGA-L and PLGA-S), BMP-2 was microencapsulated in PLGA (efficiency 80%) prior to incorporation in the polyurethane scaffold. In vitro release of BMP-2 in PBS at 37° C. was measured from 0 to 21 days by ELISA. The bioactivity of the released BMP-2 was determined by measuring alkaline phosphatase expression by MC3T3 cells incubated in released BMP-2 (FIG. 24). As shown in the Figure, the bioactivity of released BMP-2 is significantly greater than that of the negative control (no BMP-2) and less than that of the positive control (BMP-2 from the sample vial).

[0151] Below are examples of common acronyms used herein:

- [0152]** PUR Polyurethane
- [0153]** PDGF-BB Platelet-derived growth factor-BB
- [0154]** BMP-2 Bone morphogenetic protein-2
- [0155]** PLGA Poly (lactic-co-glycolic acid)
- [0156]** T Tobramycin
- [0157]** PEG Poly(ethylene glycol)
- [0158]** GF Growth factor
- [0159]** PUR/PDGF-BB Polyurethane/Platelet-derived growth factor-BB composite delivery system
- [0160]** PUR/G-PDGF-BB Polyurethane/gelatin coated PLGA Granule/Platelet-derived growth factor-BB composite delivery system
- [0161]** PUR/BMP-2 Polyurethane/Bone morphogenetic protein-2 composite delivery system
- [0162]** PUR/PLGA-L-BMP-2 Polyurethane/Poly (lactic-co-glycolic acid) large particle/Bone morphogenetic protein-2 composite delivery system
- [0163]** PUR/PLGA-S-BMP-2 Polyurethane/Poly (lactic-co-glycolic acid) small particle/Bone morphogenetic protein-2 composite delivery system